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DEVELOPMENTAL BIOLOGY OF THE AUSTRALIAN HYLID FROG *NYCTIMYSTES DAYI* (GÜNTHER)

by MARGARET DAVIES* & STEPHEN J. RICHARDS†

Summary

DAVIES, M. & RICHARDS, S. J. (1990) Developmental biology of the Australian hylid frog *Nyctimystes dayi* (Günther). *Trans. R. Soc. S. Aust.* **114**(4), 207-211, 30 November, 1990.

The Australian hylid frog *Nyctimystes dayi* (Günther) lays large unpigmented eggs that hatch no later than stage 22. Early larvae are nourished by a well-developed yolk sac and feeding commences after stage 24. Tadpoles exhibit adaptations to fast flowing streams. The mouth disc is large and sucker-like with two upper and three lower rows of labial teeth. The oral disc is hemispherical and the floor of the labrum is covered with well-developed ridges. Tadpoles can overwinter and metamorphose in the following spring/summer.

KEY WORDS: *Nyctimystes dayi*, tadpoles, life history, lotic adaptations.

Introduction

The Australopapuan hylid frog genus *Nyctimystes* Stejneger comprises species that are associated with streams in mountainous or upland regions. The sole Australian representative, *Nyctimystes dayi* (Günther), is confined to rainforest in upland northeast Queensland (Czechura *et al.* 1987).

Parker (1936) described the tadpoles of *N. cheesmanae* (as *N. montana*) and *N. semipalmata* whilst Czechura *et al.* (1987) described a tadpole of *N. dayi.* Other than these descriptions of tadpoles little is known about the life history of *Nyctimystes* species other than that tadpoles are stream-dwelling and have sucker-like mouths (Griffiths 1963; Menzies 1974; Zweifel 1983).

In the course of a study of the community ecology of tadpoles in a rainforest stream on Mt Spec, Queensland, one of us (S.J.R.), collected and reared newly-hatched tadpoles of *N. dayi.* This series was supplemented with field-collected tadpoles at various stages. Here we describe this material and comment on the behaviour of tadpoles of *N. dayi* in the stream.

Materials and Methods

Tadpoles were collected from Birthday Creek, 7 km NW Paluma, on 16.ii.1990 and reared in aerated water in 350 ml plastic containers. Larvae fed on algae provided by algae-covered rocks. Temperature of the room was maintained at 24-27°C. Specimens were preserved in 5% formalin.

Larvae were staged (where possible) according to Gosner (1960). Total length and body length (in mm) were taken using dial calipers measuring to 0.05 mm or an eyepiece micrometer.

Line drawings were made with the aid of a Wild M8 stereo dissecting microscope and attached camera lucida.

Field observations were made every fortnight; the creek was sampled intensively for tadpoles during the day and a 100 m stretch of creek was searched at night to record frog activity.

Results

N. dayi is a spring/summer breeder; calling was first heard on 29.ix.1989. Males called at night from rocks and low foliage along rapidly-flowing stretches of the creek. Calling had ceased by 1.iii.1990.

Amplexus is axillary (Fig. 1) and eggs are laid in a cohesive clump but with discrete egg capsules, under rocks in rapidly-flowing water (Czechura *et al.* 1987; pers. obs.). A clutch collected during February 1989 contained 107 eggs. Eggs are unpigmented and a sample of five eggs laid on 18.xii.1989 has a mean diameter of 2.5 mm (range 2.3-2.6 mm). Mean capsule diameter was 3.42 mm (range 3.3-3.5 mm). Only one capsule was evident.

The limbs develop within a membranous sac until they protrude from the sheath. Hence early limb bud stages of Gosner (1960) (i.e. stages 26-31) could not be judged without damage to larvae.

The earliest stage examined was stage 22, collected and preserved on 16.ii.1990 (Fig. 2). The

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Fig. 1. An amplectant pair of *Nyctimystes dayi* from Birthday Creek.

cornea is transparent and the auditory vesicle is apparent behind the eye. Ventrally the eye is unpigmented whilst the remainder of the eye is suffused faintly with pigment. Heavier coloration is concentrated postero- and anterolaterally along the dorsal surface. Two pairs of gill filaments protrude from the gill plate. The anterior gills comprise six and the posterior pair four filaments. The external nares are open and unelevated. Unpigmented adhesive organs have a flocculent appearance (Fig. 2). The transversely-oval mouth disc has formed and the mouth is open. Some ridges are apparent, precursors to the labial tooth rows and the horny beak. Labial papillae are absent. The tail is slightly curved dextrally (Fig. 2). Nutrition is supplied by a large yolk-filled body cavity. The anal tube is dextral but not open. The tail fin is transparent.

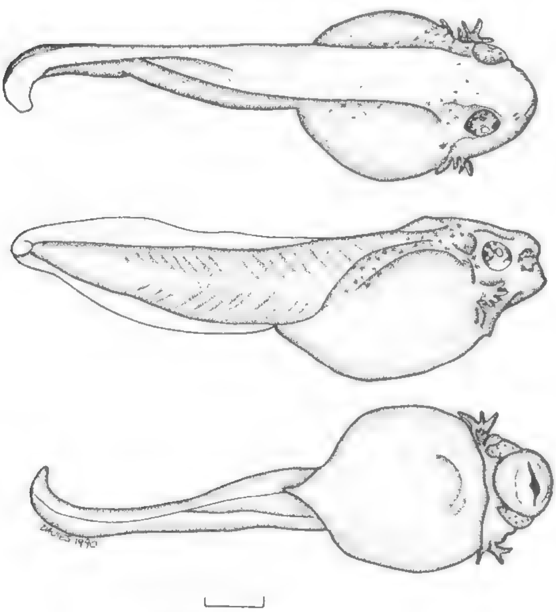

Fig. 2. Dorsal, lateral and ventral views of a newly-hatched tadpole of *Nyctimystes dayi* at stage 22 (preserved 16.ii.1990). Scale bar = 1 mm.

By 17.ii.1990 larvae were at stage 24. The external gills are covered by the operculum on the right hand side, but a small fringing of filaments remains exposed on the left hand side. The auditory vesicle is no longer detectable and the external nares are slightly elevated on stalks. Pigmentation of the eye is complete except for a ventral nick. The adhesive organs are undetectable. The precursor ridges of the horny beak and the labial tooth rows are clearly delineated and faint keratinization of the anterior upper tooth rows is apparent. Labial papillae are not formed. The dorsal fin extends from behind the head whilst the ventral fin extends posteriorly from the anus (Fig. 3). The tail musculature is faintly dusted with pigment granules anteriorly.

By 18.ii.1990, larvae were at stage 25. The gills are enclosed totally by the operculum and the spiracle has formed on the left hand side

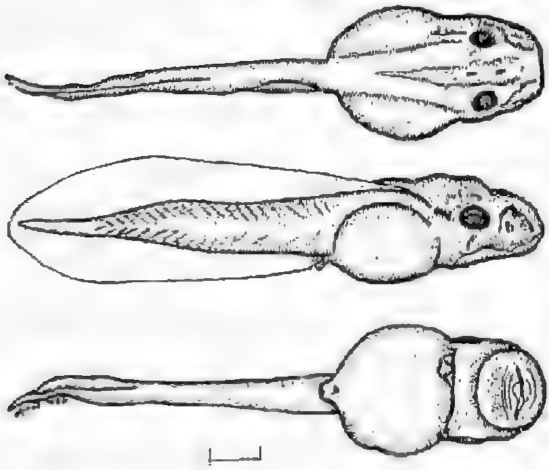

Fig. 3. Dorsal, lateral and ventral views of a tadpole of *Nyctimystes dayi* at stage 24 (preserved on 17.ii.1990). Scale bar = 1 mm.

ventrolaterally. At this stage it appears flattened and the orifice is directed posteriorly. The eye is fully pigmented. The nares are clearly elevated. Keratinization is apparent on the two upper labial tooth rows and on the horny beak. The anterior labial papillae are prominent and the ridges on the floor of the posterior lip are forming. The anal tube is open, oriented dextrally. Pigmentation is present along the dorsal musculature of the tail. The dorsal fin arises anteriorly from the junction of the body and the tail. The yolk sac is partially grooved foreshadowing the coiled gut of the feeding tadpole.

Larvae collected on 19.ii.1990 have dense body pigmentation and a light dusting of pigment in the anterior third of the tail musculature. Further tail pigmentation is confined to the dorsal extremities of the tail musculature. Two of the lower tooth rows are lightly keratinized and all labial papillae are formed. The floor ridges of the posterior lip are clearly detectable but incomplete. The spiracle remains adpressed to the ventrolateral surface of the body. The stalked nares are directed anteriorly. These larvae could not be staged because of the difficulty in locating limb buds.

Tooth rows, labial papillae and labial ridges are complete in larvae collected on 20.ii.1990. Coiling of the gut is more pronounced. The vent has moved slightly more medially and the surrounding tissue has expanded providing a sheath which protects and hides developing limb buds. The anterior attachment of the dorsal fin has moved slightly posteriorly and arises from a position slightly anteriorly from the junction of the body with the tail muscle. A faint dusting of pigment appears on the posterior extremity of the dorsal fin.

The gut is clearly coiled but remains yolk-filled in larvae sampled on 22.ii.1990. The spiracle curves posterodorsally. Although the anal tube is more medial, it still opens dextrally to the midline. The posterior lower tooth row is now complete. Pigmentation extends ventrolaterally covering portions of the yolky gut. Pigmentation is apparent on the tail fin on the medial dorsal extremities.

Dimensions of tadpoles are shown in Table 1.

A tadpole at stage 36 is illustrated in Fig. 4.

The body is widest just posterior to the eye and is broadly ovoid. The snout is evenly rounded in dorsal view and tapers to a truncated ventrally-directed upper lip. The nares are dorsolateral and elevated on tubes opening anterolaterally. The eyes are dorsolateral, moderately large and fitting snugly into the optic cup. The spiracle is sinistral, ventrolateral and not visible from above. It opens posteriorly by a narrow orifice and the diameter of the tube decreases slightly from its origin to its orifice.

The gut is coiled and the tadpole is feeding; the cloacal tube is now median. The lower limbs and developing feet protrude from a membranous sac on each side.

The tail fins are arched and rounded terminally. The dorsal fin extends for about ⅘ of the tail muscle and is deepest about ½ way along its length. The ventral fin extends from the cloacal tail piece and is deepest at its posterior ⅓. The tail musculature is deep and tapers to a fine point posteriorly.

The mouth is large, transversely oval, ventral in position (Fig. 5) and occupies the area anterior to the spiracle. Papillae surround the mouth disc. The anterior papillae are more pronounced than those laterally and posteriorly, these being little more than

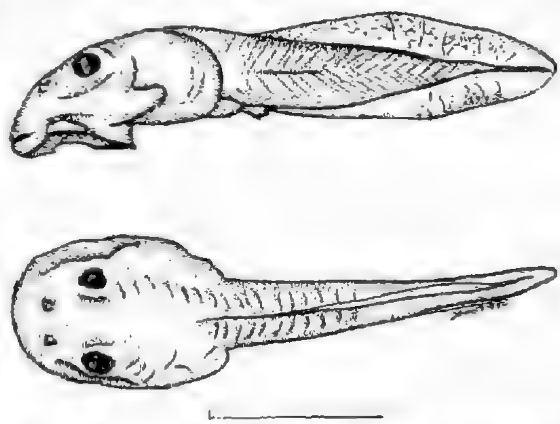

Fig. 4. Lateral and dorsal views of a tadpole of *Nyctimystes dayi* stage 36. Scale bar = 10 mm.

TABLE 1. *Body lengths of tadpoles of* Nyctimystes dayi.

Stage	Date	n	Mean body length (mm)	range	Total length
22–3	10.xi.1989	7	4.51	4.2–5.0	
25	10.xi.1989	11	7.95	7.0–8.4	
31	8.i.1990	2	14.75	14.5–15.0	
38	8.i.1990	4	15.05	14.5–15.8	
39	1.ii.1990	2	16.05	15.6–16.5	
42	1.ii.1990	1	15.6		
22	16.ii.1990	2	4.08		9.28
24	17.ii.1990	1	4.2		10.9
25	18.ii.1990	1	4.6		11.5
indet.	19.ii.1990	1	4.8		11.7
indet.	20.ii.1990	1	4.8		12.2
indet.	21.ii.1990	1	5.1		12.8
indet.	19.i.1990	1	6.2		15.0
indet.	19.i.1990	1	7.5		16.7
indet.	19.i.1990	1	7.9		19.2
indet.	19.i.1990	1	9.4		19.9
indet.	19.i.1990	1	8.7		20.9
indet.	19.i.1990	1	8.6		21.0
indet.	19.i.1990	1	8.7		21.3
indet.	19.i.1990	1	10.7		25.9
36	19.i.1990	1	12.5		32.8
37	19.i.1990	1	13.6		35.0
41	19.i.1990	1	14.3		35.6

serrations in some areas. The lateral edges of the mouth are directed dorsally (Fig. 4) (a laterodorsally curved lip). The posterior surface of the mouth disc (behind the tooth rows) is inclined posteroventrally and is ridged medially across its width. Some ridges are complete, others comprise two or three elevations. There are two upper and three lower complete labial tooth rows comprising short, very closely-applied, keratinized teeth. The horny beak is of moderate proportions.

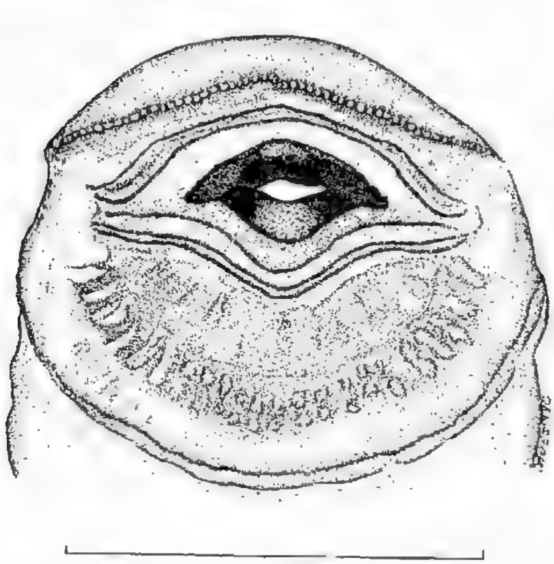

Fig. 5. Mouth disc of a tadpole of *Nyctimystes dayi* at stage 36. Scale bar = 5 mm.

The flesh of the mouth disc is translucent and unpigmented. The body is pigmented and pigmentation extends lightly along the dorsal and medial portions of the tail musculature. The remainder of the tail musculature is dusted with pigment granules. Small areas of the fins are lightly pigmented and sparse chromatophores are located elsewhere on the fins.

Tadpoles from eggs laid in early summer complete development in 3–4 months. Those eggs laid in late summer may overwinter and metamorphose the following summer (Trenerry 1988[1]; pers. obs.).

At metamorphosis one froglet measured 15.5 mm S-V and was light grey with dark flecks.

Stage 22–23 tadpoles remain attached to the egg mass under rocks in riffles and do not disperse until at least stage 24. At this stage the oral disc is well developed.

Stage 25 larvae (but with yolk still visible in the gut) were occasionally found aggregated in large numbers under single rocks. Aggregation behaviour also was observed in the laboratory: tadpoles from a single clutch aggregated in a clump which immediately reformed after disturbance. This behaviour persisted until the gut was fully formed.

In life, early stage tadpoles (stages 22–23) are bright yellow; later stages become pigmented with

[1] Trenerry, M.P. (1988) The ecology of tadpoles in a tropical rainforest stream. Honours thesis, Dept of Zoology, James Cook University of North Queensland. Unpubl.

pale brown. These tadpoles have distinct light patches on the tail.

During the day, tadpoles graze on algal-covered rocks in the fastest torrents of the stream. When disturbed they release their grip on the rocks and are swept a short distance downstream where they shelter under rocks or in rock crevices. Tadpoles are very strong swimmers, but their ability to remain in riffles even during the strongest floods (as evidenced by sampling following cyclones) probably is aided by sheltering under rocks.

Discussion

Tadpoles of *N. dayi* are adapted to fast-flowing mountain streams. The ventral, suctorial mouth-discs, the narrow tail fins and the ventrolateral spiracular opening are characteristic of species occurring in torrent environments (Duellman & Trueb 1986). The development of the larval gut parallels that described in the sympatric *Litoria eucnemis* by Davies (1989), but *N. dayi* exhibits more extreme adaptations to the lotic life style.

The tooth row pattern is similar to other hylids (Martin & Watson 1971), although the undivided nature of all the tooth rows is unusual amongst Australian hylids, being shared by *L. lesueuri* which also shows lotic adaptations (Martin & Watson 1971).

The median vent found in later-stage tadpoles is shared by congeners but is unusual amongst hylids (Martin & Watson 1971). The covering by a membranous sac of the developing hind limb buds is presumably an adaptation to protect these structures. This feature has been observed in the limnodynastine *Mixophyes* spp. (Watson & Martin 1973; Davies in press) and *L. nannotis* (pers. obs.). Larvae of these species are lotic and found in fast-flowing streams.

Acknowledgments

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THE NATURE AND INCIDENCE OF POST-AXIAL, SKELETAL ABNORMALITIES IN THE FROG *NEOBATRACHUS CENTRALIS* PARKER AT OLYMPIC DAM, SOUTH AUSTRALIA

by JOHN L. READ* & MICHAEL J. TYLER†

Summary

READ, J. L. & TYLER, M. J. (1990) The nature and incidence of post-axial, skeletal abnormalities in the frog *Neobatrachus centralis* Parker at Olympic Dam, South Australia. *Trans. R. Soc. S. Aust.* 114(4), 213-217. 30 November, 1990.

Samples of 315 specimens of the frog *Neobatrachus centralis* from four sites at Olympic Dam, South Australia, included 12 specimens exhibiting skeletal abnormalities of the limbs. Examination revealed a predominance amongst the abnormal specimens of partial or complete ectrodactyly, most commonly involving terminal components of the fourth toe. The overall incidence of abnormalities is comparable to those occurring at undisturbed sites in other countries.

Radionuclide levels in tadpoles from the sampled sites were very low or not detectable, and were not associated with the incidence or nature of the abnormalities there.

KEY WORDS: Skeleton, abnormalities, *Neobatrachus centralis*, radionuclides.

Introduction

Olympic Dam Operations (O.D.O.) manages a large copper-gold-uranium-silver mine at Olympic Dam, approximately 500 km north of Adelaide. Mining commenced in 1984 and the metallurgical plant there started production in August 1988.

Airborne, aquatic and biotic environments are monitored stringently for both conventional (e.g. SO_2, SO_3 and total particulates), and radioactive emissions by Olympic Dam Operations, in accordance with the South Australian Government approved Environmental Management Program (Roxby Management Services 1986), and the Waste Management Program, Olympic Dam Project (O.D.P. 1987). The plant and animal communities in all habitats in the Olympic Dam region are monitored regularly to determine the possible effects, if any, of the mining and processing operations on species diversity, abundance and condition. There have been no measurable effects on the environment, outside the immediate vicinity of the metallurgical plant, that can be attributed to the mining or processing operations (O.D.P. unpubl.).

In addition to the general monitoring program, more detailed studies are conducted on certain indicator organisms, such as frogs, to enhance the sensitivity of the monitoring program. Frogs are very sensitive to radiation (Emery & McShane 1980) and have proved to be useful indicators of radioactive emissions (Nishimura 1967; Tyler 1989) and trace elements (Browne & Dumont 1979). Frogs are also the most common vertebrate animals associated with claypans: regions of natural heavy metal and radionuclide accumulation. A photograph of one of the claypans at Roxby is presented by Tyler (1989, plate 3).

Following rains in the semi-arid Olympic Dam region, the frog *Neobatrachus centralis* is exceptionally common adjacent to claypans and flooded swales (O.D.O. unpubl.). The ease of capture and identification of physical abnormalities in live specimens makes it an ideal subject as a potential indicator of environmental emissions at Olympic Dam.

Here we document an initial survey of frog deformity levels at control sites where there are negligible emission levels, and at sites in close proximity to the metallurgical plant where emission levels, while remaining very low, are detectable (O.D.O. 1989[1]).

Materials and Methods

On 12.iv.1989, approximately 80 tadpoles were collected from two water bodies within 1 km of the Olympic Dam mine and metallurgical plant (Claypan, ENW5), and from a pond 16 km south of the mine (EV 308) (Fig. 1). The EV 308 site is near a continuous radionuclide and airborne emission monitoring site. It has not detected any emissions from the mine, and hence is a valid control site.

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[1] Olympic Dam Operations (1989) Environmental Radiation Monitoring Annual Report. (Report to the S.A. Govt) Unpubl.